ON THE BIOLOGY OF CNEPHIA ORNITHOPILIA AND SIMULIUM VERNUM (DIPTERA: SIMULIIDAE) IN INSULAR NEWFOUNDLAND

AUGUSTINE NKADI OKAEME

ON THE BIOLOGY OF *Cnephia ornithophilia* and *Simulium vernum* (Diptera: Simuliidae) IN INSULAR NEWFOUNDLAND

by

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TABLE OF CONTENTS

ABSTRACT

The habitat and life history of Cnephia ornithophilia, Davies, Peterson and Wood; and Simulium (Eusimulium) vernum (Macq) were investigated in 49 streams in order to characterize the habitat and larval distribution.

Cnephia ornithophilia larvae were restricted to pond outlets feeding in small permanent streams in forested areas. The distribution of larvae indicated a relatively broad range of conditions in terms of stream width, depth, current velocity and substrate choice. This species is univoltine, and eggs begin to hatch in September. There are eight larval instars with a prolonged period of development (September to May). The larvae formed clumps, were sedentary, and relocate to the undersurface of substrates in winter, while physiologically the growth rate between instars is highly variable and they have a wide thermal tolerance (0-30^{0}C).

Simulium vernum larvae are more widely distributed in streams than C. ornithophilia, occurring both at pond outlets and elsewhere in temporary and permanent streams both in forested and barren areas. These streams ranged from small tricklets to streams up to 3 m wide. The local species is cytotype "B" and bivoltine with the first generation from May to July and the second generation July to August.

Using emergence and bantam-baited traps it was found that C. *ornithophilia* emerges in May, only during the day (1500 -- 1900 h) when stream temperature is 10-12°C. Host seeking females were anautogenous limited to the forest and between 1300 - 2000 h. Oviposition apparently occurs within pond near the outlet, based on location of first instars.

Simulium vernum emerges in June (1100 - 1900 h), and in August (0900 - 1300 h), for the first and second generation respectively. Host seeking females are anautogenous with activity within forest and forest fringes between 9000 - 2000 h. The prevalence of *Leucocytozoon* in females was 19.04%, with a high attack and blood feeding rates, indicating a good vector. Oviposition is along streams near sunset (1700 h).

Although the seasonal occurrence of C. *ornithophilia* and S. *vernum* is later and prolonged in Newfoundland, they are ecologically similar to populations occurring elsewhere on the continent.

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LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

The study of ornithophilic blackflies in Canada gained impetus when they were recognized as pests (Shewell 1955), and vectors of avian hematozoa (Fallis et al 1956, Bennett and Fallis 1960), certain filarioid nematodes of ducks (Anderson 1956) and viruses (EE), (Anderson et al 1961).

The literature on the family Simuliidae is vast and topics such as ecology, bionomics, disease transmission, host seeking behaviour and control strategies have been well summarized by Laird (1981). The taxonomy has also been studied, including both morphological (Puri 1925a,b, Snodgrass 1944, Peterson and Dang 1981) and cytological characterization of some species (Rothfels 1979, 1981). Important taxonomic works which deal mainly with species occurring in eastern North America include Davies et al (1962), Stone (1963), Stone and Snoddy (1969) and Peterson (1981). In Newfoundland, Lewis (1973) studied the distribution and bionomics of blackflies on the island and identified eleven ornithophilic species. Cnephia ornithophilia Davies, Peterson and Wood was not recognized in this particular study due to uncertainty of the identification at that time. However, Lewis and Bennett (1973), under the designation Simulium (Eusimulium) latipes (auct. nec Meigen) and recognized by

Crosskey and Davies (1972) as Simulium (Eusimulium) vernum (Macq), stated it was an important bird feeder.

The first specimen of C. ornithophilia in North America was collected in 1949 (Davies et al 1962); subsequently its blood feeding habit was studied by Bennett (1960) who referred to it as Cnephia "U". Procunier (1975a,b) first reported the existence of C. ornithophilia in Newfoundland, based on cytological characterization of the larvae. Recently he traced the phylogenetic relationships between Cnephia species based on cytological evidence (Procunier 1982a,b). The ecology of Cnephia ornithophilia has been little studied except for some observations of its life cycle and rearing in the laboratory (Tarshis 1972). The only studies in Newfoundland have been in a brief description of its stream habitat (Colbo 1979) and a laboratory study on competition for space with Prosimulium mixtum (Harding and Colbo 1981).

Studies on Simulium vernum include taxonomy (Crosskey and Davies 1972), distribution (Davies et al 1962, Wood et al 1963), feeding behaviour and ovarian development (Davies and Peterson 1956, Pascuzzo 1976), and vector potential (Bennett 1960, Bennett and Fallis 1960, Bennett and Coombs 1975).

The objects of this study were:

I. Characterization of habitat and larval distribution of the two species, C. ornithophilia and S. vernum.

II. Life cycle studies including larval growth pattern, number of generations, emergence pattern and temperature tolerance of C. ornithophilia and S. vernum.

III. The characterization of adult host-seeking behaviour.

IV. Associations of larvae of winter developing simuliid species at pond outlets.

MATERIALS AND METHODS

Study areas

The study areas lie between $48^{o}7.5'$; $48^{o}49'$ East latitude and $54^{o}9'$ $54^{o}45'$ North longitude on the Avalon Peninsula of Newfoundland. The geology is mainly bedrock of precambrian sediment and lower palaezoic strata (Dawson 1963) with predominantly acidic volcanic rocks and intrusive granite batholiths (Keat 1970). Water drainage systems generally show a broken profile with ponds, bogs and swamps connected by stream segments. The peninsula has a maritime climate (Banfield 1981) strongly influenced by the interaction of the Gulf Stream, North Atlantic Drift and Labrador current, resulting in frequent rain, snowfall and fog. The prevailing surface wind is north westerly produced by the north Icelandic current and the west high pressure of Hudson Bay. The wind direction and velocity are highly variable. High wind velocity (in excess of 30 km/h) was commonly noted in exposed areas. The westerly wind is also influenced by the east spring-ward movement (Banfield 1981) of anticyclonics in spring and northward extension of Bermudian sub-tropical anticyclonic in summer.

During the winter of 1981 and 1982 ponds froze over in late December to January with ice break up occurring in April. Flooding occurred when ice and snow melted.

The principal vegetation zones are woodland, composed mainly of <u>Picea mariana</u> (black spruce), <u>Abies balsamea</u> (balsam fir), <u>Larix</u> spp (Larch), <u>Betula papyrifera</u> (White birch), and bog (essentially <u>Chamaedaphne-Sphagnum</u>), in poorly drained areas. The central portion of the Avalon is tundra-like barrens covered with low grasses, sedges, mosses and lichens, interspaced with primarily ericaceous shrubs. In areas disturbed by wood cutting, fire, dams, buildings, bridges and highway construction the vegetation is open with succession of grasses and shrubs.

<u>Cnephia ornithophilia</u>

<u>Field studies of pre-imaginal stages</u>

Larval distribution of <u>Cnephia ornithophilia</u> as well as associated species, was investigated by selection of 3-5 natural substrates at each study site. Larvae were preserved in 70% alcohol, for later identification and counting to determine larval density. Pond outlets and downstream areas from which larvae were absent were sampled on more than one occasion. The choice of pond outlets was limited by accessibility but an effort was made to sample pond outlets in forest, open barren and disturbed areas.

At each sampling site the current velocity in m/sec was measured with a current meter (Ott® propeller driven). Substrate types, stream depth in cm, and width in meters were recorded at points of collection of larvae. Water analyses were carried out in a number of streams during the year. In Hugh's and Healey's Pond outlets, attempts were made to study the distribution of larvae on substrates, by recording the number of larvae on the sides or under the stones.

A study on the vertical distribution of _C. ornithophilia_ on fixed 15 cm square cement blocks was conducted at two sites at Oxen Pond outlet. Distribution of larvae along the depth of the block was determined by counting the number of larvae within the interval 1-5 cm, 6-10 cm and 11-15 cm measuring from the top of the block. Observations were from December to May.

Larval distribution along the stream at Oxen Pond outlet was monitored twice monthly by taking the mean densities of four samples collected in each of the four marked zones of the stream as shown in Table 1. Measurement of larval density was continued until pupation. Observations on aggregation or clumps attachment of _C. ornithophilia_ were made.

<u>Laboratory studies on pre-imaginal stages</u>

Larvae collected from the field were identified using a Zeiss dissecting microscope at magnification of 10X - 40X, and the taxonomic keys of Davies _et al_ (1962),

Table 1.

Characterization of Oxen Pond outlet (October 1981)

Site	Distance from outlets (m)	Depth (cm)[a] Range	Depth (cm)[a] Mean	Width (cm)[b] Range	Width (cm)[b] Mean	Current velocity[c] m/sec mean	Predominant Substrate[d]	Bank	Stream Character	Remark
1	0-20	21-41	35	150-300	200	0.30	Grass, branches of shrubs, few boulders	Spruce forest vegetation on side with grass and bog on one side	Shallow slow channel	Partially shaded with growth of vegetation in channel flowing through bog *C. ornithophilia* present
2	21-60	42-48	45	30-120	70	0.38	Grass, few rocks, few boulders	- do -	Deep defined channel but with fast flow in constrictions	Partially shaded with deep channels in bog *C. ornithophilia* present
3	61-80	12-26	20	10-140	100	0.08	Rock, some grass	Spruce forest on both sides	Wide shallow, sluggish flow	Pool, some tire and can dumping - ve
4	81-100	12-18	15	50-100	40	0.35	Rock, boulders, twigs	Spruce forest on both sides	Shallow, fast flow with riffle	Shaded, *C. ornithophilia* present
5	140-200	25-70	50	30-120	50	1.08	Boulder, twigs, grass	Completely enclosed in spruce forest on both sides	Deep, very fast flow with riffle	Shaded, very fast and steep riffle - ve

a b c d = based on five different measurements

Wood et al (1963), Stone and Snoddy (1969), Merritt et al (1978) and Peterson (1981).

To determine the number of instars, forty-seven first instar larvae were collected from Oxen and Left Pond outlets and reared individually in 25 ml test tubes at 15 + 1°C. A current was produced by bubbling air through the tubes. Larvae were fed Tetra R fish food at a rate of 0.05 mg/litre/2 days. After each moult the exuvial head capsule was recovered and mounted on a slide. The maximum width of the cephalic apotome of each recovered head capsule was measured in micrometers. The number of instars prior to pupation was noted for each larva by counting the number of head capsules produced.

Temperature tolerance of early instars (3-4) of Cnephia ornithophilia was studied by subjecting them to temperatures of 15° C, 20 °C, 30°C, 40°C, 50°C and 60°C. Four replicates of thirty larvae in 250 ml distilled water were set at each temperature in an adjustable mechanical shaker (50-150 RPM). Larvae were fed on Tetra R fish food at a rate of 0.5 mg/litre/48 h. Mortality of larvae during each temperature treatment was recorded after 96 h.

Various studies on larvae were conducted using an artificial stream consisting of plexiglass troughs, the first 150 cm x 2.5 cm x 10 cm and the second 150 cm x 9 cm x 10 cm. A continuous flow of water was produced by means of a water pump. A water reservoir containing 45 litres aged tap water supplied the pump. Stones and pebbles were arranged along the trough to act as substrates and to vary water velocity. Overall current velocity was regulated by varying the angle at which the trough stood using a rectangular wooden wedge, 2.5 cm x 4 cm and by adjusting the pump overflow. Calibration of rate of flow was determined by measuring the amount of water flowing through the trough in ml/sec at the outflow using a 250 ml measuring cylinder and timer.

Temperatures were controlled by incorporating a refrigerating coil with a thermostat (-20°C - +10°C) into the 45 litre reservoir of the artificial stream system. The copper cooling coils were placed in two glass cylinders to prevent their contact with water flowing into the trough to prevent any larval mortality due to copper toxicity. An adjustable stirrer, 50-500 rpm, was incorporated in the reservoir to facilitate uniform heat exchange and to prevent settling of food particles. Water in the reservoir was changed weekly.

Following establishment of known rates of water flow and temperature in the stream system the following two studies on <u>Cnephia ornithophilia</u> larvae were conducted. (i) The effect of temperature on larval distribution on stone substrates; (ii) aggregation of larvae.

Adults

An emergence trap was used to assess the emergence pattern of adult Cnephia ornithophilia. The trap consisted of a 30 cm square cage covered by small mesh netting, with the open end facing the surface of water. The traps were fixed at Oxen and Left Pond outlets over large populations of pupae. The number of flies emerging hourly was monitored for six consecutive days. The wing length of collected flies was measured. The sex and physiological age of females were determined by noting the stage of development of the ovarioles. Mating behaviour was looked for in the field and in the laboratory. Success in mating was determined by dissection of the female spermathecae. The emergence of field collected pupae was studied in the laboratory at 18°C and light regime of 12 h light: 12 h dark for six consecutive days.

The seasonal occurrence of adult flies was studied by use of a portable miniature suction trap based on CDC mosquito trap. It consisted of a 1250 rpm motor, 10 cm propellor, 6 volt battery as power source, an aluminium cylinder 15 cm high and 15 cm diameter with a collecting net at the base. A small piece of dry ice in a small container was attached to the cylinder to provide a source of carbon dioxide (Fig. 1).

In another investigation the apparatus developed by Bennett (1960) for trapping ornithophilic simuliids

Figure 1.

Miniature suction carbon dioxide baited trap.

(Fig. 2) was used to study the attraction of flies to a live bantam chicken bait. Trapping was attempted at all hours of the day but later limited to 1400 h to 2000 h. The choice of trapping sites was by trial and error. Sites tested included rocky outcrops, coniferous forest (open or completely forested), open stream sides, and low shrub at Oxen Pond Botanic Park. Subsamples of captured flies were dissected to study stage of ovariole development, presence of spermatozoa, and Leucocytozoan sporozoites in the salivary glands.

Simulium vernum

Preimaginal field studies

Field observations on the distribution of S. vernum in the streams were made with notes on habitat character. Seasonal occurrences of the various stages of development were noted.

Adult

Trapping studies were conducted in the same manner as for Cnephia ornithophilia. These studies included seasonal occurrence, emergence pattern, host seeking activity, presence or absence of parous flies, Leucocytozoan infection and mating behaviour. Supplementary sweepnet collections were also made along Oxen and Left Pond outlets to investigate ovipositing behaviour.

Figure 2.

Bantam baited ornithophilic trap at trapping site.

RESULTS AND DISCUSSION

Cnephia ornithophilia

Stream Habitat

Larvae were found to occur within the first 100 m along the stream below pond outlets (Table 2). In outlets where larvae occurred, they were always present within the first 50 m at some period during the year but their occurrence in the 55-100 m zone was variable. Larvae were usually not encountered beyond 100 m. Larval densities diminished downstream (Table 3). In most cases, larvae concentrated at obstructions in areas of fast flowing water. Colbo (1979) previously noted the occurrence of C. ornithophilia larvae in pond outlets in the St. John's area.

Distribution of larvae in relation to width, depth and current velocity at pond outlets showed larvae were found in streams with widths of 0.3-6.5 m (mean width 2.06 m, Table 4), with 73% of observed positive samples being in streams of 1.1-3.0 m wide. They occurred at depths of 6-60 cm, (mean depth 28.5 cm, Table 5), with most larvae within 21-60 cm depth. The current velocity was 0.12-1.08 m/sec (mean 0.42 m/sec) with 80% of observed larvae occurring at 0.6-1.08 m/sec current velocity (Table 6). Cnephia ornithophilia has a wide tolerance to width, depth and current velocity which agrees with the observations of Merritt et al (1978). However, in insular

Table 2

Occurrence of <u>Cnephia ornithophilia</u> larvae along the stream to show limited outlet distribution (Percentage probability in bracket)

	Site and date	0	5	10	20	25	30	48	50	55	60	65	70	74	75	80	81	90	100*	120	150	200	250+
1	Healey's Pond																						
	19 Sept. 1981	X	X		X	X			X														
	2 March 1981				X	X		X			0		0										
2	Left Pond																						
	23 Sept. 1981	X	X	X		X												0	0		0		0
	2 Oct. 1981	X	X	X		X												0	0				
	19 Feb. 1983	0	X		X				X						X	0		0	0	0	0		0
3	Maggotty Pond																						
	21 Oct. 1981			X		X			X										0				
4	Robins Pond																						
	17 Dec. 1981	X		X	X				0														
5	Oxen Pond																						
	14 Sept. 1981	X	X	X		X			X		0					0	X		0				
	19 Sept. 1981	X	X	X		X			X		0					0	X		X				
	2 Oct. 1981	X	X	X		X			X		0					0	X		0	0	0		
	19 Feb. 1983	0		0		X			X		0					0	X		0		0		
6	Picco Pond South																						
	17 Dec. 1981	X	X						X	X	X	0		0									
7	Mt. Scio bog																						
	17 Dec. 1981	X	X						X	X		0		0									
8	Buttler Park Pond																						
	26 Nov. 1981					X			X		X	0	0		0								

Table 3

Relative larval density/cm^2 of Cnephia ornithophilia as it changes with time downstream of Oxen Pond outlet.

Dates	Sites		
	0-20 m	21-60 m	61-92 m
August 11/8/81 26/8/81	-ve	-ve	-ve
September 2/9/81 27/9/81	4.08	0.47	-ve
October 2/10/81 29/10/81	66.67	200.00	-ve
November 1981	68.90	29.17	0.33
December 1981	75.30	29.00	0.44
January 28/1/82	Frozen	Frozen	-ve
February 2/2/82	Frozen	Frozen	0.11
March 11/3/82	-ve	0.17	0.23
April 22/4/82	0.030	0.29	0.56
May 22/5/82 31/5/82	Pupae	0.49 + Pupae	0.54
June 3/6/82 12/6/82	Pupae	Pupae + larva	Pupae
July 22/7/82	Pupae	Pupae + larva	Pupae

Table 4

Occurrence of Cnephia ornithophilia larvae in relation to width (in meters) of streams

Width Range (m)	0.10-1.00	1.10-2.00	2.10-3.00	3.10-4.00	4.10-6.50	6.6-10.00
No. of samples	41	15	8	1	8	10
No. with larvae	13	9	4	1	6	0
Percentage	31.7	60	50	100	75	0

Table 5

Occurrence of *Cnephia ornithophilia* larvae in relation to depth (in cm) of streams

Depth range (cm)	0-20	21-40	41-60	61-80	81-above 90
No. of samples	38	20	16	3	6
No. with larvae	16	9	8	0	0
Percentage	33.3	45	50	0	0

Table 6

Occurrence of *Cnephia ornithophilia* larvae in relation to current velocity (in m/sec) of streams

Velocity range (m/sec)	0-0.5	0.6-1	1.1-1.6	1.7-2.2	2.2-3.5
No. of samples	33	10	2	2	4
No. with larvae	17	8	0	0	0
Percentage	51.5	80	0	0	0

Newfoundland larvae occurred mainly in small streams with a narrower range of physical characteristics, than in Michigan where they occurred in large streams with widths up to 10 m (Merritt et al 1978).

Larvae were found to attach to available substrates. In early fall (September), most pond outlets had abundant trailing vegetation with attached larvae. As winter progressed and vegetation decreased, larvae were found particularly on the under surfaces of stones and boulders. They were attached to substrates with little or no algal growth while susbtrates covered with algae had no larvae. However, there was no preference for any particular substrate type including cans, tires, broken bottles and debris. *Cnephia ornithophilia* larvae were noted to occur in low light-zones (6-200 lux), in culverts and under bridges as well as in open channels.

Analyses of water from outlets and corresponding downstream sections in fall and summer for pH, O_2, PO_4 and hardness did not indicate any differences between streams and sections of streams that correlated to the distribution of larvae. The ranges in selected chemical characteristics were: pH - 5.6-6.2; oxygen - 2.5-4.0 mg/litre; phosphate 0.05 mg/litre; and hardness 14.4-15.2 Ca^{++} Mg^{++}) mg/litre (Table 7). The range of pH and dissolved oxygen levels of streams with larvae in insular Newfoundland agrees with the pH and O_2 ranges found by Tarshis (1973).

Table 7

Selected water analysis of pond outlets and downstream sections

Pond	Site (m)	pH Range	Oxygen (mg/l)	PO_4 (mg/l)	Hardness Ca^{++} Mg^{++} (mg/l)	Season
Left pond	0-20	5.6-5.8	2.5	0.05	15.2	early fall, September 1981
	80-100	5.8-6.0	2.5	-	-	early fall, September 1981
Mt. Scio (Higgins)	0-25	6-6.2	3.9	0.05	14.4	early fall, September 1981
	50-10	5.8-6.2	3.9	0.05	15.0	early fall, September 1981
Healey's Pond	0-25	6-6.2	3.9	0.05	15.2	early fall, September 1981
	50-70	5.8-6.0	4.0	0.05	15.2	early fall, September 1981
Power Pond	0-25	5.8-6.0	2.5	0.05	15.2	early fall, September 1981
	50-80		3.9	0.05	15.2	early fall, September 1981

(Cont'd)

Table 7 (Cont'd)

Pond	Site (m)	pH Range	Oxygen (mg/l)	PO_4 (mg/l)	Hardness Ca^{++} Mg^{++} (mg/l)	Season
Oxen Pond	0-10	5.6-6	3.26	0.05	15.0	Summer, July 1982
	11-20	5.8-6	3.26	0.05	15.2	Summer, July 1982
	21-60	5.4-6	3.26	0.05	10.4	Summer, July 1982
	100-110	5.6-6.6	2.26	0.05	8.2	Summer, July 1982

Distribution of larvae in local stream system

Many stream systems in insular Newfoundland possess a broken profile interrupted by ponds, small lenthic bodies, and sections of steep gradient (Yoxall 1981). *Cnephia ornithophilia* is restricted to pond outlets, but larvae were not found in outlets of ponds in completely barren surroundings (Table 8). One major factor influencing which outlets were colonized was the presence of forest habitat in the vicinity of the stream (Table 8).

Seasonal occurrence

First instar larvae were observed in pond outlets from mid-September until late November. They grew throughout the winter as shown by the increases in widths of their cephalic apotomes (Fig. 3). However, samples collected in September had a greater variation in cephalic apotome size than samples collected later in the fall or winter (Fig. 3). Stream temperatures observed during larval development were 0-18°C, with 18°C occurring during early September and lowest temperatures observed from January to March. The period of larval development on the island is 8-9 months (September to May). It is a longer period of development than recorded elsewhere, for example: Maryland, 5-6 months (November to April), Tarshis (1973); Michigan, 2-3 months (February to April), Merritt *et al* (1978). The prolonged period of larval

Table 8

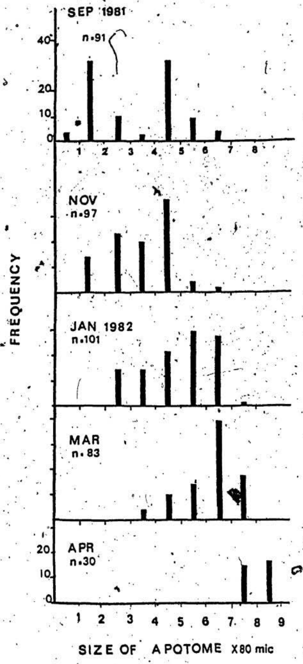
The relationship between forestation and distribution of simuliid species at the Pond outlets (percentages in bracket) 1981-1982

	Area Surrounding Pond			Outlet Area		
Species and no. of Ponds	Completely forested	Partial forested	*Barren (> 200m)	Completely forested	Partial forested	Barren (> 50m)
No. of sampled sites	19	11	19	8	15	26
C. ornithophilia	12(63.16)	7(63.64)	0(0)	6(75)	11(73.30)	4(15.20)
P. mixtum	14(73.70)	7(63.60)	9(47.40)	6(75)	12(80)	11(42.30)
St. mutata	6(31.60)	2(18.20)	2(10.50)	5(62.50)	6(40)	2(7.60)
S. vittatum	5(26.30)	3(27.30)	6(31.60)	7(87.50)	7(46.70)	4(15.20)

*Barren area with nearest vegetation 200 m around pond and 50 m at outlet

Figure 3.

The growth of Cnephia ornithophilia larvae from Oxen Pond as indicated by apotome widths. (Scale numbers x 80 um = width).



SEP 1981
n.91
NOV
n.97
JAN 1982
n.101
MAR
n.83
APR
n.30
FREQUENCY
40
20
10
0
1 2 3 4 5 6 7 8 9
SIZE OF APOTOME X80 mic

development in insular Newfoundland is probably due to the longer period of low stream temperatures resulting from the prolonged winters and cool springs (Banfield 1981). Mansingh et al (1972) and Colbo and Porter (1981) have shown that lower temperatures reduce the rate of larval development.

Number of instars

Through rearing larvae individually in the laboratory it was determined that larvae passed through eight instars. Within each instar there was considerable size variation (Table 9, Fig. 4). The growth ratio between moults was 1.19-1.82 with an overall average of 1.42. Cephalic apotome measurements for larvae of any one instar overlap considerably with measurements of larvae in adjacent instars. Clear peaks of cephalic apotome width classes corresponding with instars can only be seen for the first 3 or 4 instars. Previous attempts to determine the number of larval instars in the closely related C. dacotensis by Ross and Merritt (1978) using measurements of cephalic sclerites were unsuccessful.

Maximum size increase occurred between the last two instars (Table 9). Studies on growth pattern in a winter species, Prosimulium mixtum, associated the maximum growth in late instars with the influence of temperature (Merritt et al 1982).

Table 9

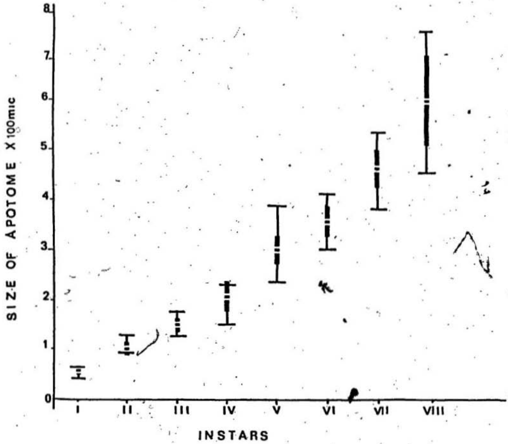
Cnephia ornithophilia maximum cephalic apotome width (mm) based on laboratory reared larvae to show the eight instars and growth pattern

Instar	Number of larvae	Mean $\bar{x}$	(S . D)	Range	Mean increase between instars a	Growth ratio between moult b
I	9	0.055	(0.008)	0.038-0.61		
					0.045	1.82
II	9	0.100	(0.015)	0.088-0.123		
					0.048	1.48
III	16	0.148	(0.012)	0.123-0.176		
					0.059	1.40
IV	18	0.207	(0.030)	0.152-0.236		
					0.093	1.45
V	16	0.300	(0.031)	0.258-0.380		
					0.056	1.19
VI	10	0.356	(0.030)	0.304-0.410		
					0.104	1.29
VII	6	0.460	(0.038)	0.402-0.532		
					0.138	1.30
VIII	12	0.598	(0.087)	0.456-0.742		

$a = \bar{x}_{inst+1} - \bar{x}_{inst}$; $b = \dfrac{\bar{x}_{inst+1}}{\bar{x}\ inst}$

Figure 4.

<u>Cnephia ornithophilia</u> mean cephalic apotome width based on laboratory reared larvae. (Scale x 100 m = width).



SIZE OF APOTOME X 100mic
0
1
2
3
4
5
6
7
8
I
II
III
IV
V
VI
VII
VIII
INSTARS

A similar plasticity of growth pattern to that shown by _C. ornithophilia_ has been noted in a closely related species, _C. dacotensis_ (Ross and Merritt 1978). Procunier 1982b) noted that B-chromosomes in these species can exert an effect on their growth and should thus be examined further.

Morphological differences observed between instars were slight but some generalities can be stated as follows:

INSTAR I: Scleritization of head capsule highly reduced; egg burster present, situated at the axis of V-shaped dorsal sclerite; scleritization along lateral wall with a very deep post-genal cleft (epicranial cleft) extending almost to hypostomium.

INSTARS II-IV: Head capsule completely scleritized and cephalic apotome clearly visible; three groups of hypostomal teeth, two lateral and 5-7 central serrations; head spot visible but not delineated clearly.

INSTARS V-VIII: Hypostomal teeth less distinct; head spots visible, but sizes and pigmentation of spots inconsistent, thus not useful as characters separating instars; late instars with clearer suture line along cephalic apotome; imaginal discs representing pupal respiratory filament buds, leg buds, and wing buds visible.

The general morphology of *C ornithophilia* larvae agreed with the description of Merritt *et al* (1978); however, there were slight differences in the color of larvae. On the island, larvae had brown to reddish brown intersegmental bands and dark brown head capsules, with weakly defined head spots. Michigan larvae (Merritt *et al* 1978) had greyish intersegmental bands, light brown head capsules and clearly defined head spots.

Behaviour of larvae in stream

Cnephia ornithophilia larvae were found individually or in clumps (Fig. 5). Larval density on randomly selected natural substrates from 15 pond outlets between September 1981 to May 1982, ranged from 0.009-5.60 larvae per cm^2 (mean 2.60 larvae/cm^2, n = 40). However, in Oxen Pond outlet, densities were considerably higher (Table 3).

Larvae attach to substrates by first sticking small pads of silk to the surface of the substrate and then grasping the pad with their anal hooks. In time, small particles adhered to the silken pad, giving it a brownish appearance. The association between larvae within clumps was very close with little spacing between individuals but their anterior ends were free to enable filtering activity. Clump sizes varied from few to several hundred larvae attached to interconnected masses of silk.

Figure 5.

Aggregation of Cnephia ornithophilia larvae.

In September, when early instar larvae first appeared in streams, they occurred on trailing vegetation near the surface, but as winter approached and the stream temperature lowered, larvae were found more frequently on the under surface of stones. Field experiments on depth performance indicated that more larvae occurred 11-15 cms deep than in the upper 0-10 cms (Table 10).

Another experiment using natural stone substrates was conducted at Healey's and Hugh's Pond outlets. Larval distribution was recorded on randomly selected stones of sizes 28-660 cm^2. It was found that out of 988 C. ornithophilia collected, 85.6% (846) were under stones (Table 11). A non-parametric sign test revealed this difference was significant, $Z = 1.64$ ($P < 0.05$), thus indicating the more frequent occurrence of larvae under surface of stones. These results confirm Tarshis's (1973) observation that larvae move under substrates in winter. The movement could be triggered by lower stream temperature as suggested by Tarshis (1973). Under stone habitat in winter may protect larvae against ice formed by supercooling and ice movement.

Co-occurring simuliid winter species

During field sampling three simuliid species were found co-occurring in the same streams with Cnephia ornithophilia. These were Prosimulium mixtum, Stegopterna

Table 10

Distribution of simuliid larvae with depth at Oxen Pond outlet

Month	Date	No. larvae and depth below surface (in cm.)								
		C. ornithophilia			P. mixtum			St. mutata		
		0-5	6-10	11-15	0-5	6-10	11-15	0-5	6-10	11-15
December	9-12-81	38	15	56	0	50	23	0	0	0
		0	2	9	0	0	0	0	0	0
December	17-12-81	26	25	42	15	3	9	0	0	5
		6	9	156	18	13	17	0	0	0
January	3-1-82	1	8	13	15	6	8	3	0	4
		8	0	17	10	0	0	17	0	5
February	2-2-82	12	0	25	10	0	0	0	0	0
					0	0	15	3	0	5
May	11-5-82	25	0	15	14	15	3	0	0	0
		35	25	0	2	10	3	2	0	0

(Cont'd)

Table 10 (Cont'd)

		No. larvae and depth below surface (in cm.)								
Month	Date	C. ornithophilia			P. mixtum			St. mutata		
		0-5	6-10	11-15	0-5	6-10	11-15	0-5	6-10	11-15
Total		154	86	358	84	97	77	25	0	19
mean		15.40	8.60	35.80	8.40	9.70	7.70	2.50	0	1.90
Percentage %		25.80	14.40	59.80	33.10	38.20	28.70	56.80	0	43.20

Table 11

The association of winter simuliids C. ornithophilia (co),
S. vittatum (sv), P. mixtum (pm), St. mutata (st) on stone substrates

Site	No. of samples	Total larval density (Larvae/cm^2)	No. of larvae on stone Understone co	pm	st	sv	Side of Stone co	pm	st	sv
Hughes Pond outlet	7	.046	130	30	9	45	35	101	9	33
Healey's Pond outlet	12	.20	293	168	18	407	36	295	3	149
Total	19		423	198	27	452	71	396	12	182
		z =	7.38	1.50	1.28	3.20				

z = non-parametric sign test value 1.64 at 95 level significance

35

mutata and Simulium vittatum. Like C. ornithophilia, most species occurred over a wide range of stream width, depths and current velocities. However, St. mutata occurred in a narrower range of current velocity. These species, unlike C. ornithophilia, occur in streams with or without surrounding forest (Table 8). These findings generally agree with those of Colbo (1979).

The frequency of co-occurrence of the winter species is summarized in Table 12. It was common for two or more winter species to occur in the same stream. In Healey's and Hugh's Pond outlets, C. ornithophilia and S. vittatum larvae were found more frequently on the under surfaces of stones (Table 11) while P. mixtum and St. mutata larvae were more frequently on the sides of stones. The seasonal appearance of first instar larvae in streams differ between the winter species. First instar C. ornithophilia appeared in mid-September; P. mixtum in late September (29/9/81 - 10/11/81) with an extended period of hatching; S. vittatum in late October (4/10/81 - 20/10/81) and St. mutata in late December. The seasonal distribution of these winter species, other than C. ornithophilia, agreed with Lewis and Bennett (1974).

Artificial stream experiments

Distribution of larvae on substrates showed that some larvae occurred under stones regardless of temperature (Table 13), although a higher percentage of

Table 12

Simuliid species association in

forty-nine pond outlets during the winter

Species association types	No. of sites	Frequency %
A = C. ornithophilia	0	0
B = P. mixtum	11	22.24
C = St. mutata	0	0
D = S. vittatum	3	6.12
A + B	1	2.04
A + D	2	4.08
B + C	1	2.04
B + D	1	2.04
A + B + C	6	12.24
A + B + C + D	4	8.16
A + B + D	6	12.24
No larvae found	14	28.84

Table 13

Relative distribution of Cnephia ornithophilia on stone substrates in an artificial stream with respect to temperature.

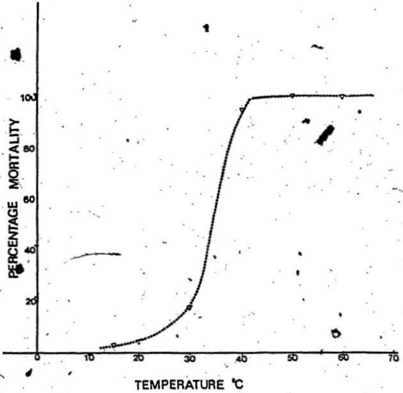
Temperature °C	Time spent (hrs.)	Number of observations	Mean larval population/250 cm^2 stones: Understone	+ S D	Top of stone	+ S D
20°	48	4	35.25	8.98	23.75	5.65
15°	48	4	38.75	18.49	14.75	6.34
10°	48	4	53.35	15.02	6	3
5°	48	4	69.75	8.75	7	2.29

larvae occurred under stones at lower temperatures. This distribution supports the observation of larvae relocating under stones during winter as noted in this study and reported by Tarshis (1973).

The temperature tolerance curve (Fig. 6) showed that C. ornithophilia larvae had an increase in mortality rate with increasing temperature. Larvae tolerate 30°C (Fig. 6) but at 50°C death was instantaneous. This tolerance of warm temperatures differs from other winter occurring simuliid species. Tarshis (1973) reared C. ornithophilia at 25°C, while P. mixtum survived only below 20°C (Mokry et al 1981). Stegopterna mutata is very sensitive to heat and has to be reared at 10°C (Mokry 1978, Mokry et al 1981). Colbo and Porter (1981) reared S. vittatum at 15°C and at 25°C but flies developed from larvae reared at 25°C were smaller and less fecund. This wide temperature tolerance of C. ornithophilia may permit survival in a wide range of stream conditions and could account for the wide north American distribution. The distribution of C. ornithophilia extends from Texas and South Carolina, north to the Canadian Prairies and into the boreal forest of Ontario (Stone and Snoddy 1969, Tarshis 1973, Procunier 1982a) and Atlantic Canada.

Figure 6.

Cnephia ornithophilia larval mortality rate at various temperature in the laboratory.



PERCENTAGE MORTALITY
100
80
60
40
20
0
10
20
30
40
50
60
70
TEMPERATURE °C

Pupae

Pupae were first sighted in the field in early May and persisted over a period of three weeks. The pupal cocoon is loosely woven and poorly formed, as observed by Tarshis (1973). Pupae have 30-45 branched respiratory filaments. Because of the fragility of the cocoon, pupae were collected from the field by removing pupae with their substrates to avoid damage.

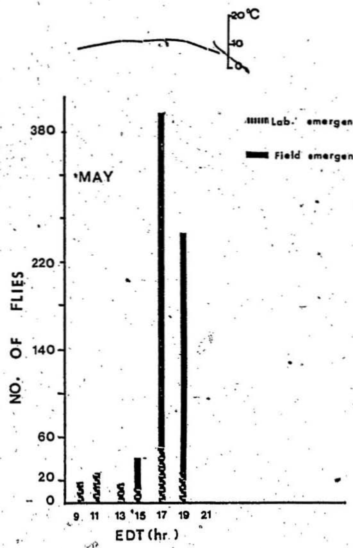
Pupae occurred from 0-100 m from pond outlets and their distribution did not differ from that of late instar larvae. Although a few individual pupae were found on the upper surfaces of substrates, large clumps of pupae were found on the under surfaces. Pupae showed no obvious orientation in relation to direction of stream flow such as observed in other simuliids (Burton 1966, Disney 1969).

Emergence pattern and size of flies

Cnephia ornithophilia adults were first collected from the field in emergence traps in mid-May (19/5/82). Emergence pattern shown in Figure 7 was based on results of six consecutive trapping days (20-25th May 1982). Adult emergence was between 1500 h to 1900 h, peaking at 1700 h (Fig. 7). The mean sunrise and sunset hour was 0415 h and 1941 h respectively. No flies were collected before 1500 h or after sunset. The stream temperature recorded during periods of emergence was

Figure 7

Diurnal emergence pattern of Cnephia ornithophilia. Water temperatures shown at top of figures.



20°C
10
0
Lab. emergence
Field emergence
MAY
380
220
140
60
20
0
NO. OF FLIES
9
11
13
15
17
19
21
EDT (hr.)

10-12°C. A total of 637 flies were collected with a sex ratio of 1 female: 0.55 male. The sex ratio agrees with that observed by Tarshis (1973). Under laboratory conditions at 18°C, 12 h dark: 12 h light, emergence occurred only throughout the light period.

Flies emerging from Oxen and Left Pond outlets differed in size, based on wing length. Flies from Left Pond outlet were larger with a wing length of 3.30-3.80 mm with a mean of 3.44 ± 0.02 mm for females and 2.45-3.55 mm with a mean of 2.75 ± 0.05 mm for males compared to 1.99-3.33 mm with a mean of 2.90 ± 0.06 mm for females and 2.08-2.57 mm with a mean of 2.40 ± 0.2 mm for males from Oxen Pond outlets. The differences were significant for both sexes (F_1 66.19, F_2 121.99, $P < 0.01$). Differences in sizes of blackflies have been attributed to nutritional availability and temperature (Scriber and Slansky 1981, Colbo 1982). The productivity of different ponds is known to vary widely in Newfoundland (Kerekes 1975). Since *C. ornithophilia* larvae probably feed from seston flowing from ponds, larvae in different outlets may received varied levels of nutrition which affect adult size.

Activity and Habitat

The adult fly activity (May to August) based on bantam-baited trap (Table 14) was later than recorded in Ontario (Davies *et al* 1962) and in Maryland (Tarshis

Table 14

Total number of host-seeking simuliids

captured in bantam-baited trap 1982

Species	Month and no.				Total	Total Catch success	Percentage Blood fed
	May	June	July	August			
C. ornithophilia	19	68	2	-	89	11.66	-
S. vernum	-	124	342	20	485	63.56	64.54 (313)
P. mixtum	10	83	18	-	111	14.55	-
S. venustum/verecundum	-	9	13	7	29	3.80	3.44
S. vittatum	-	-	4	-	4	0.52	-
St. mutata	2	1	2	-	5	0.66	-

1973). Trapping with bantam-baited traps was most productive in forest habitats (Table 15). In a total of 28 sampling days, 82.4% flies caught were in forest mid-canopy and 17.6% in open wooded areas. No flies were captured in grass and shrub, barren or rock out-crops habitat (Table 15). These findings indicate C. ornithophilia is associated with forest habitat, and agrees with Bennett (1960) who suggested mid-canopy preference.

Trapping of C. ornithophilia was attempted throughout the day. It was found that activity of the flies started gradually at 1300 h with greatest activity between 1600 - 1800 h (Fig. 8). Bennett (1960) considered the fly as a late evening feeder but findings herein (Fig. 8) suggest host seeking activity over a longer period than previously reported. Flies were not captured in an unbaited miniature carbon dioxide trap (Fig. 1) placed in presumably suitable sites.

Flies that were blood fed, parous or naturally infected with Leucocytozoon were not caught in bantam-baited traps. Attempts to induce blood feeding in the laboratory using live bantam chicken were unsuccessful. C. ornithophilia did not feed on bantam chicken under trapping conditions, nor were they observed feeding in traps baited with several duck species (Tarshis 1973). However they feed on sylvatic birds (Bennett 1960), indicating some host preference. Host preference by

Table 15

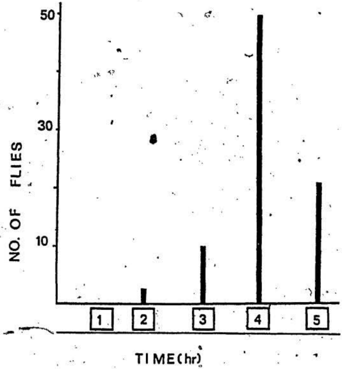
Habitat selection of <u>Cnephia</u> <u>ornithophilia</u>
based on catches from bantam-baited traps

Habitat	No. of trapping days	No. flies
Forest habitat	16	89
Open grass and shrub Mt. Scio	4	0
Outcrop, open rock habitat with mosses Mt. Scio	3	0
Open woodland with shrub near Left Pond	5	19
Total	28	108

Figure 8

Diurnal activity of host-seeking <u>Cnephia</u> <u>ornithophilia</u> based on catches from bantam-baited trap, with the numbers representing periods of activity.

1. 0900 h to 1200 h
2. 1300 h to 1400 h
3. 1500 h to 1600 h
4. 1700 h to 1800 h
5. 1900 h to 2000 h



50
30
10
NO. OF FLIES
1
2
3
4
5
TIME(hr)

blackflies has been reported in several ornithophilic species (Davies and Peterson 1956, Bennett 1960, Lowther and Wood 1964, Bennett and Fallis 1971).

Neither oviposition or mating was observed in the field or among captured flies in the laboratory. However, female flies collected from bantam-baited traps had spermatophores or sperm cells in the spermathecae. The ovaries of nulliparous females held with a sugar source in the laboratory did not develop beyond stage II. Although ovipositing C. ornithophilia were not observed, based on the location of its first instars, one would conclude that oviposition occurs in ponds within forested areas. The collection of first instar larvae in the slow flow section where water leaves the pond, strongly suggests eggs are deposited within the pond near its outlet.

Trap collection of flies during periods of high wind (20-30 km/h) or at ambient temperatures below 10°C, or above 24°C was reduced, and no activity was observed during rain.

Cnephia ornithophilia in insular Newfoundland compared to other areas of North America

Some general observations on the biology of C. ornithophilia are summarized in Table 16. Cnephia ornithophilia in insular Newfoundland is univoltine, overwintering in the larval stages. This life cycle pattern

Table 16

Some ecological and morphological characteristics of Cnephia ornithophilia in North America

Character	Newfoundland	Ontario	Maryland	Alabama	Michigan
A. Larvae		1	2	3	4
Seasonal occurrence	Sept. to May		Nov. to April	Jan. - March	March - April
Generation	Univoltime		Univoltime	Appear univoltine	
Number of instars	8				
Larval general appearance	Uniform light brownish to reddish brown head capsule, lighter brownish dark but weakly defined head spots				bicoloured, greyish intersegmental bands, contrasting greyish brown to reddish brown body. Head spots clearly marked with strong contrasting dark colour
Location along stream	Limited to outlets < 100 m	along stream	stream runoff		
Physical character of stream					
Range of depth	6 - 60 cm				10 - 150 cm
Range of width	0.3 m - 6.3 m		3 - 10 m (large)		3 - 10 m (Large)
Range of current velocity	0.12 - 1.08 m/sec		0.17 - 1.12 m/sec		
Temperature	0.18°C	15.5°C (60°F)	0.5°C - 15°C		
pH	5.4 - 6.2		6.9 - 7.4		
Oxygen	2.5 - 4.0 mg/l		10 - 13 ppm		6 - 10 mg/litre
Substrates	trailing vegetation rock, boulders, object		trailing vegetation, objects		trailing vegetation, rock
Surrounding vegetation	forest and woodland only.		appears forested		marsh and lowland brush
Nature of stream	permanent	permanent	permanent		permanent
Larvae understone migration	understone migration as winter approaches		larvae moves beneath debris 0.5°C - 11°C		

Table 16 (cont'd)

Character	Newfoundland	Ontario	Maryland	Alabama	Michigan
B. Pupae					
Seasonal occurrence	May		December to April		April to July
Location in stream	outlets in aggregation no obvious arrangement		along stream sparsely arranged		along stream sparsely arranged
Cocoon	loosely formed; naked pupae only in laboratory; brownish to reddish colour. Has two short dorsal spines.		Loosely woven with particles of silica incorporated; few naked pupae in field.		Well formed defined anterior margin. Terminal abdominal segment with two short dorsal spines.
Respiratory filament	5 main branches 30 - 45				Variable arise from 5 main branches,
C. Adults					
Seasonal occurrence	May to July	May to June	March to May	Feb. to May	April to May
Emergence period	1500 h to 1900 h				
Wing size (mm)	female 3.33 - 3.8 male 2.45 - 3.55	4.2 - 5			
Female	large, dark brown; leg lighter yellowish; claws with basal tooth	dark brown to blackish-brown; legs yellowish to brownish to orange brown; claw with basal tooth			
Male	Small dark brown; macrotrichia of wing hairlike and spiniform; short calcipala; terminalia hairy with dark, large tapering distimere with one apical tooth.		Known but undescribed		
Habitat	Sylvatic, woodland tree canopy 3-5 m	sylvatic[6]	open area and woods		

Table 16 (Cont'd)

Character	Newfoundland	Ontario	Maryland	Alabama	Michigan
Diurnal Activity	1300 - 2000 h	evening[6]			
Host		woodland birds[6]	birds	flickers	
Vector potential		high	Laboratory transmission[7] of <u>Leucocytozoon</u>		

1. Davies <u>et al</u> 1962
2. Tarshis 1973
3. Stone and Snoddy 1969
4. Merritt <u>et al</u> 1978
5. Procunier 1982a
6. Bennett 1960
7. Tarshis 1976
8. Tarshis and Adkins Jr. 1971

agrees with records from other areas of the continent (Tarshis 1973, Merritt *et al* 1978). The larvae pass through 8 instars in Newfoundland and that is the only study in which the number of larval instars has been determined. The period of development is longer in Newfoundland than elsewhere (Table 16) which is attributed to the prolonged winter and delayed spring. While larvae from all areas are morphologically similar, the brownish head capsule and weakly defined head spots of the Newfoundland population contrasts with the light colour and dark headspots of larvae from Michigan.

Distribution of larvae along streams has not been defined in other areas of the continent. However, in Newfoundland *C. ornithophilia* does not occur beyond 100 m from pond outlets, concentrating within the first 50 m. The association of some simuliid species with outlets has been attributed to effects of food resources (Carlsson *et al* 1977, Wotton 1979). *Cnephia ornithophilia* larvae remained in the pond outlets throughout development. It is assumed larval location in outlets is due to the proximity of ponds, and their use made by larvae of seston originating from the pond. Most seston is recruited from the pond and probably steadily decreases in quality and quantity downstream (Sheldon and Oswood 1977).

In insular Newfoundland, larvae occur in small streams in forested areas. The distribution in Michigan is in larger streams in areas of low land brush and woods

(Merritt et al 1978). The difference in distribution pattern may be influenced by adult fly activity. In insular Newfoundland the host-seeking adults were captured in the forests, as in Ontario (Bennett 1960) but adult activity in other areas is unknown.

Cnephia ornithophilia larvae differ from the other local winter-developing species in such features as high temperature tolerance, and variable growth rate and clump formation. Tarshis (1973) in Maryland reported migration of larvae during winter to the undersurface of substrates, confirmed in the present study.

The pupae occur in loosely formed cocoons (Tarshis 1973) and possess a variable number of respiratory filaments arising from 5 main branches (Merritt et al 1978). It was observed that pupae occur in clumps under stones. They lack the orientation to the stream flow characteristic of pupae of other simuliid species (Burton 1966).

The adult females of C. ornithophilia have been described by Davies et al (1962) and Stone and Snoddy (1969), but the male is undescribed. In insular Newfoundland the males can be recognized by: size 2.08-3.55 mm, brown macrotrichia, and hairlike spiniform of wing, short calcipala, terminalia hairy with a dark large tapering distimere with only one apical tooth.

Host seeking females are anautogenous. They are active from mid afternoon to late evening within forest areas and appeared to have preference for sylvatic birds (Bennett 1960). Oviposition is presumed to occur at pond outlets based on the location of the first instars. There is no data on the method of oviposition, so it is not known whether it follows pattern known for other Cnephia species. The methods used by females of genus Cnephia include: deposition of eggs on substrates near the surface of water, crawling below the surface after which ovipositing flies die as in C. pecuarium (Bradley 1935) or striking the abdomen on the water surface while in flight, depositing eggs into the water as in C. dacotensis (Davies and Peterson 1956). Mating of C. ornithophilia does not occur at emergence sites as in C. dacotensis (Twinn 1936, Hocking and Pickering 1954) and probably requires some swarming flight as in many simuliid species (Downes 1969).

Simulium (Eusimulium) vernum

Habitat and distribution of larvae and pupae along streams

Simulium vernum larvae were found throughout the stream system in channels 0.10-3 m wide temporary or permanently flowing, and mostly at depths of 4-30 m. The current velocity where larvae were found was 0.05-2.43 m/sec. Larvae were found mostly on the surface of trailing vegetation and, in contrast to C. ornithophilia, appear to tolerate algal growth as they were found on substrates covered with algae. Stream temperatures recorded during studies were 5°-18°C. Pupae were found in as wide a range of habitats as the larvae. Cocoons were small (2-3 mm) and characterized by a single hook-like projection at the anterior end and four unbranched respiratory filaments. The pupae were found both on the surface and under trailing vegetation with the anterior opening facing downstream. The wide rage of habitat distribution of S. vernum may explain the island wide distribution reported by Lewis and Bennett (1973). However it differs sharply with the distribution of C. ornithophilia found in forested areas (Table 2, 17) due to C. ornithophilia adult forest activity.

Seasonal occurrence and species complex

First instar larvae were observed in early May, with pupae occurring in early June and adults 7-10 days later to complete the first generation. The second generation first instar larvae were observed in early July at Oxen Pond outlet, Pickavance Creek and Left Pond outlet. They pupated in early August and adults emerged 7 days later to complete the second generation.

The appearance of first instars in the spring (May) indicated S. vernum overwintered in the egg stage as in Ontario (Davies et al 1962) but in Quebec the larval stage overwintered (Wolfe and Peterson 1959). The Newfoundland population may differ from those in mainland Canada, where both univoltine and multivoltine populations occur (Davies et al 1962, Imhof and Smith 1979). Both

Table 17

Characteristics of some streams with <u>Simulium</u> <u>vernum</u>

Stream	Nature of flow	Width m	Depth cm	Current velocity m/sec	Substrate	Surrounding
Oxen Pond						
site I	Permanent	1.20	6	0.28	trailing vegetation	forested
II	Permanent	0.50	10	0.39	trailing vegetation	forested
III	Permanent	0.50	30	0.075	trailing vegetation	forested
IV	Permanent	3	30	0.58	trailing vegetation	forested
Benthobservatory bog	Temporary	0.30	4	2.43	grass	grass
Power Line						
(Trans Canada) I	Permanent	0.90	10	-	rock	barren
II	Permanent	-	-	-	rock	barren
Mt. Scio runoff	Permanent	0.70	6	0.16	vegetation boulders	forested

larvae of the first and second generations herein were identified as cytotype "B" by K.H. Rothfels and C. Brockhouse (Per. Comm.). However at least three sibling species designated "A", "B" and "Labrador", based on cytological differentiation, occur in Canada (Rothfels and Brockhouse, Per. Comm.). The "A" and "B" cytotypes are widespread in Canada while the "Labrador" cytotype is known only from Labrador. The discrepancy in the number of generations between regions and the diversity of cytotypes suggest S. vernum is a complex of species but in insular Newfoundland one bivoltine species exists.

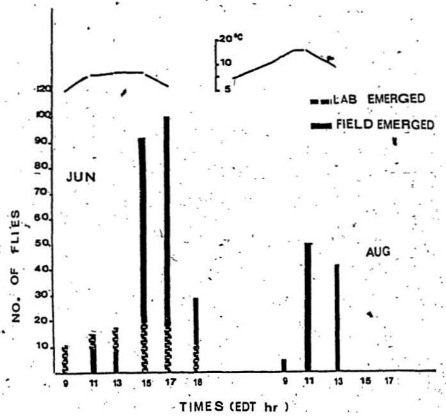
Adult

Emergence pattern

The first generation (June) of S. vernum emerged between 1100 h to 1900 h with peak period at 1500 h to 1700 h. No flies emerged before 0800 h, and during darkness. The stream temperature recorded during periods of emergence was 10°C-12°C and mean sunrise and sunset hour (12-17th June 1982) was 0401 and 2002 h respectively. A total of 244 flies were collected with a sex ratio of 1 female: 0.43 male. Male flies were the first to be collected in emergence traps. For the second generation (August), emergence was between 0900 h to 1300 h with a peak at 1100 h. Flies were not collected after 1400 h (Fig. 9), and during darkness. The stream temperature

Figure 9

Diurnal emergence pattern of first and second generation of *Simulium vernum*. Water temperature shown at top of figure.



20°C
10
5
LAB EMERGED
FIELD EMERGED
JUN
AUG
NO. OF FLIES
120
100
90
80
70
60
50
40
30
20
10
9
11
13
15
17
18
9
11
13
15
17
TIMES (EDT hr)

recorded during emergence was $10^{o}-18^{o}C$ and mean sunrise and sunset hour was 0402 h and 1911 h respectively (7-12th August 1982). A total of 98 flies were collected with a sex ratio of 0.45 female: 1 male. In the laboratory at 12 h light: 12 h darkness at $18^{o}C$, emergence occurred only during the light period. Temperatures of $10^{o}C$ and above were not recorded in the stream before 1300 h in early summer but occur before 1000 h in late summer (August). These findings suggest that the pattern of emergence may be influenced by the interaction of stream temperature and light.

<u>Adult behavior</u>

Adult flies trapped at Oxen Botanic Park were identified by B.V. Peterson as <u>Simulium vernum</u>. It was found during studies that the adult flies differed slightly from published descriptions. The second antennal segment of females was longer instead of being shorter than the third segment, compared to <u>S. vernum</u> (<u>S. latipes</u>) described by Davies, <u>et al</u> (1962).

Adult female <u>S. vernum</u> were caught in forest and forest fringed habitats. Activity of flies based on bantam-baited traps showed the activity occurred from June to August (Table 14). Daily activity in the first generation started mid-morning (0900 h) with optimal host seeking activity 1700–2000 h (Table 18). Seventy-three

Table 18

Diurnal activity of the first and second generation of Simulium vernum.

First generation - 16 collecting days, June 16 - August 2;

Second generation - 5 collecting days, August 16-29.

Activity	0900-1200	1300-1400	1500-1600	1700-1800	1900-2000	Total
First Generation						
No. of flies	34	24	70	152	204	484
% of total catch	7	5	14.4	31.3	42.2	100
No. blood fed	13	9	30	103	155	310
% of total blood fed	4.2	2.9	9.7	33.2	50	100
Second Generation						
No. of flies	-ve	-ve	-ve	4	26	30
% of total catch	-ve	-ve	-ve	9.9	90.1	100
No. blood fed	-ve	-ve	-ve	2	13	15
% of total blood fed	-ve	-ve	-ve	13.3	86.7	100

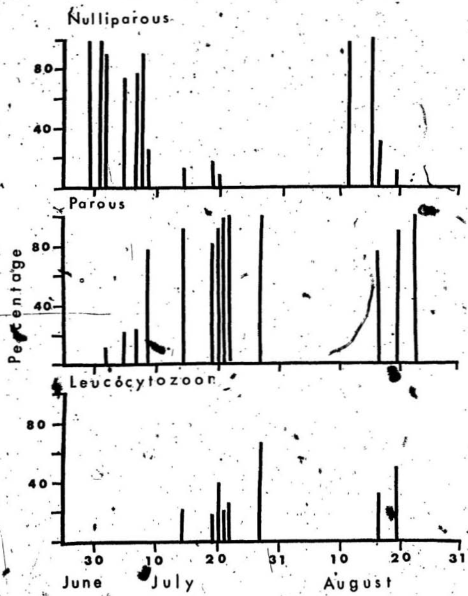
percent of the total flies caught were taken near sunset with activity extending past sunset (1945–1917 h). The temperature recorded at trapping sites when flies were active was 12°–24°C. The second generation flies were active between 1700–2000 h (Table 18). The daily activity was limited; however, it is not clear whether it was a shift of activity due to seasonality or a sampling effect due to the small population. The temperature at trapping sites when flies were active was 15°–25°C. The attack rates were higher than previously reported on the island (Bennett and Coombs 1975). The high proportion of engorgement of flies agrees with Bennett (1960) in similar studies at Algonquin Park, Ontario.

The percentage of parous females increased over the period of trapping reaching peaks in mid July for the first generation and late August for the second generation (Fig. 10).

The occurrence of Leucocytozoon sporozoites in the first generation was 14% and 22% for the second generation with overall prevalence of 14% (Fig. 10). The prevalence of Leucocytozoon was much lower in flies than the 82% occurrence reported in Newfoundland passeriform birds (Bennett and Inder 1972, Bennett and Cameron 1974). The high prevalence of Leucocytozoon in the avifauna has been attributed to the vector efficiency of S. vernum (Bennett and Coombs 1975).

Figure 10

Distribution of parous and multiparous Simulium vernum of the first and second generation.



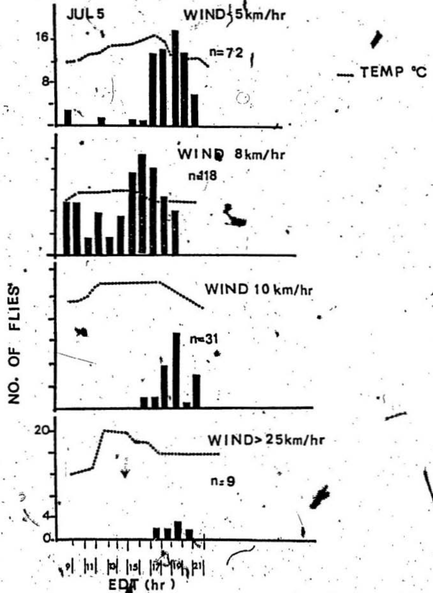
Nulliparous
80
40
Parous
80
40
Percentage
Leucocytozoon
80
40
30
10
20
31
10
20
31
June
July
August

Ovipositing females with mature eggs were caught in sweep net collections in the evenings (July 29th and August 25th, 1982), along a stream 48-50 m from a pond outlet. On August 25th, the temperature at time of collection was 16°C, 1900 h, not windy and few clouds. The time of oviposition agrees with Imhof and Smith (1979) who observed oviposition near sunset.

Some quantitative observations on the effect of weather were made on S. vernum activity. High wind reduced activity. Trapping was most successful on days of low wind (5 km/h), temperature above 10°C, and high humidity but no precipitation (Fig. 11). The numbers of S. vernum captured increased on days following periods of high wind, precipitation or low temperature below 10°C. It would be unreasonable to generalise on the effect of weather on activity of flies because of the masking effects of various environmental conditions. However, the observations herein agrees with Fallis (1964) that extreme conditions of high wind and temperature affect flight and host seeking activity of S. vernum. The relationship between temperature and activity agrees with that reported by Taylor (1963), whereby activity gradually increases with increase in temperature up to an optimum.

Figure 11

Host-seeking activity of <u>Simulium vernum</u> and effect of wind speed and temperature.


JUL 5
WIND 5km/hr
n=72
TEMP °C
WIND 8km/hr
n=18
WIND 10 km/hr
n=31
WIND > 25km/hr
n=9
NO. OF FLIES
16
8
20
4
0
9
11
13
15
17
19
21
EDT (hr)

SUMMARY

1. The distribution of larvae of Cnephia ornithophilia and Simulium vernum was characterized in 49 streams on the Avalon Peninsula, Newfoundland.

2. Cnephia ornithophilia occurred in pond outlets in forested areas. Along the stream, larvae were restricted to within 100 m of an outlet with highest concentrations of larvae within the first 50 m. They remained at the outlet throughout their development. The size of the streams ranged from 0.3-6.5 m wide, 6-60 cm in depth, with a current velocity from 0.12-1.08 m/sec.

3. Simulium vernum larvae were not restricted to streams in forested areas, occurred both at outlets and downstreams sections and in temporary tricklets or permanently flowing streams. The streams ranged from 0.10-3 m wide, 4-30 cm depth and current velocity from 0.05-2.43 m/sec.

4. Cnephia ornithophilia is the only member of the genus in insular Newfoundland. It is univoltine and has eight larval instars with a prolonged period of development (September-May). Morphologically the larvae are similar to those described elsewhere except the head spots are weakly pigmented.

5. Simulium vernum overwinters in the egg stage with the local population being bivoltine, the first generation occurring from May to July and the second generation July to August.

6. Observations in the field and laboratory showed that *C. ornithophilia* larvae readily formed clumps. They relocated to the under surface of substrates during the winter and attached to those substrate with little or no algal growth. This species has a variable growth rate between larval instars and has a wide thermal tolerance of 0-30°C.

7. The pupa of *C. ornithophilia* has a loosely formed cocoon with 5 main branches and 30-45 respiratory filaments. They occur in clumps below pond outlets with no obvious orientation to direction of stream flow.

8. *Simulium vernum* pupae are distributed throughout the stream system. They have well formed, small (2-3 mm) cocoons, with a characteristic anterior projection and four unbranched respiratory filaments.

9. Adults of *C. ornithophilia* emerged in May between 1500 - 1900 h when stream temperatures were between 10-12°C. There was significant differences in size of flies emerging from different pond outlets.

10. The emergence of adult *S. vernum* was in June for the first generation between 1100 - 1900 h when stream temperatures were 10-12°C. For the second generation, adult emergence in August was between 0900 - 1300 h when stream temperatures were 10-18°C. In laboratory experiments at 18°C, using pupae of *C. ornithophilia* and *S. vernum*, emergence occurred only during daylight hours.

11. *Cnephia ornithophilia* is anautogenous with host seeking activity limited to forests between 1300 - 2000 h.

12. *Simulium vernum* is anautogenous with a wider range of habitat than *Cn. ornithophilia*. Host-seeking activity was between 0900 - 2000 h. A prevalence of 14% *Leucocytozoon* infection in the salivary gland was found, indicating this species is a good vector of this genus of parasite in the study area.

13. Oviposition site for *C. ornithophilia* is at the pond outlet, based on the location of first instars. *Simulium vernum* oviposited along the stream near sunset (1700 h).

14. The biology of *C. ornithophilia* in Newfoundland is similar to that of this species in other regions of North America, differing in their occurrence in smaller streams and the prolonged development of the larval stages due to the early winter and delayed spring in insular Newfoundland.

15. The biology of *S. vernum* is comparable to those in other areas of the continent but the number of generations is limited to two.

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